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**Serum levels of magnesium, calcium, and phosphorus in female beef cattle (*Bos indicus*)
at a farm in San Borja, Beni, Bolivia**

**Niveles séricos de magnesio calcio y fósforo en hembras bovinas de carne (*Bos indicus*),
en un establecimiento de San Borja, Beni, Bolivia**

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Article Data

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Abstract

Macrominerals are essential for the health and productive performance of cows. In order to establish the reference ranges of serum Mg, P and Ca, in selected beef bovine females (*Bos indicus*), in San Borja, Beni, Bolivia, by age group and reproductive condition, 173 blood samples were taken. The serum analysis of Ca and Mg was determined by the end-point colorimetric method, and the P by the end-point ultraviolet method, the chemical analyzer STAT FAX 1908 PLUS was used, accuracy quality control was applied. The type of research was observational cross-sectional. Statistical analysis was performed using Microsoft Excel 2010 and InfoStat 2020. The data were analyzed with measures of central tendency, kurtosis and asymmetry coefficient, Gaussian bell and ANVA with 95 % reliability. The values of Ca, Mg and P show an almost normal Gauss curve. For Mg, a single range of 1.5 to 3.3 mg dL⁻¹ is declared (p>0.05). In P, two range values are declared per age group, (p<0.05), 4.5 to 8.6 mg dL⁻¹ for bovine females from 2 to 4 years of age and 3.9 to 8.2 mg dL⁻¹ from 5 to 12 years of age. For Ca, a single range (p>0.05) of 7.4 to 12.6 mg dL⁻¹ is declared. These results provide useful local parameters for the evaluation of mineral status, diagnosis of subclinical deficiencies, reliable interpretations and nutritional management of cattle in the region.

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Resumen

Los macrominerales son fundamentales para la salud y rendimiento productivo de vacas. Con el objeto de establecer los rangos de referencia de Mg, P y Ca séricos, en hembras bovinas de carne (*Bos indicus*), seleccionadas, en San Borja, Beni, Bolivia, por grupo etario y condición reproductiva, se tomaron 173 muestras de sangre. El análisis en suero de Ca y Mg se determinó por método colorimétrico de punto final, y el P por el método ultravioleta de punto final, se utilizó el analizador químico STAT FAX 1908 PLUS, se aplicó control de calidad de exactitud. El tipo de investigación fue observacional transversal. El análisis estadístico se realizó mediante Microsoft Excel 2010 e InfoStat 2020. Los datos se analizaron con medidas de tendencia central, curtosis y coeficiente de asimetría, campana de Gauss y ANVA con un 95 % de confiabilidad. Los valores de Ca, Mg y P muestran una curva de Gauss casi normal. Para Mg se declara un solo rango de 1.5 a 3.3 mg dL⁻¹ (p>0.05). En P se declara dos valores de rango por grupo etario, (p<0.05), 4.5 a 8.6 mg dL⁻¹ para hembras bovinas de 2 a 4 años y 3.9 a 8.2 mg dL⁻¹ de 5 a 12 años. Para Ca se declara un solo rango (p>0.05) de 7.4 a 12.6 mg dL⁻¹. Estos resultados aportan parámetros locales útiles para la evaluación del estado mineral, diagnosticar deficiencias subclínicas, interpretaciones confiables y el manejo nutricional del ganado bovino en la región.

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Introduction

In Bolivia, the Beni department has a cattle population of approximately 3,088,148 head in different categories. The extensive cattle farming system employed, despite its shortcomings, produces high-quality beef compared to other regions of the country. The beef produced is based on inexpensive, natural pastures, highly palatable, and highly nutritious, without the addition of fertilizers, herbicides, or any other harmful compounds that could affect human health¹.

In beef cows of the *Bos indicus* type, the measurement of serum levels of Magnesium (Mg), Phosphorus (P) and Calcium (Ca) acquire special relevance in tropical plains, where the mineral quality of forages can be variable and metabolic adaptation is a key factor to maintain productive balance under nutritional stress².

Mineral metabolism is an important factor in production and reproduction. Inorganic P, Ca, and Mg are the most important macronutrients. Their serum concentrations not only indicate nutritional balance but also the animal's physiological capacity to maintain mineral homeostasis despite varying management circumstances and physiological status³.

Ca is the most abundant mineral in the body, with 98 % found in bones and teeth⁴. It is of great importance in bovine metabolism, as it plays regulatory roles in blood coagulation, nerve transmission, and muscle contraction, in addition to its structural role in bone tissue. In adult cows, its plasma concentration normally ranges between 9 and 10 mg dL⁻¹, being somewhat higher in calves⁵.

P is the second most important macronutrient for bone tissue production⁶, playing an essential role in cell signaling, energy metabolism (ATP), and is a

component of nucleic acids^{5,7}. It is essential and is estimated to be the third most expensive component in an animal's diet⁷. Lack or deficiency of P in cattle causes decreased appetite, low pregnancy rates, reduced growth rate, weight loss, decreased milk production during lactation, and an uncontrolled appetite (pica), characterized by chewing bones in the field⁸.

Mg, an essential intracellular cation, functions as a cofactor in enzymatic reactions in major metabolic pathways. Extracellular magnesium is essential for muscle function, nerve transmission, and bone matrix production; it is also involved in maintaining the balance between calcium and phosphorus. Its concentration in bovine plasma tends to range between 1.8 and 2.4 mg dL⁻¹. Maintaining the normal concentration of this mineral in plasma depends almost entirely on the intake of magnesium through food⁵.

Given the importance of these minerals in reproductive and metabolic processes and overall health, there is a need to evaluate serum levels of Mg, P, and Ca in order to establish a reference range for the metabolites analyzed in beef cattle (*B. indicus*) under grazing conditions. The data obtained will provide information for diagnosing subclinical deficiencies, providing reliable interpretations, optimizing nutritional management, improving reproductive efficiency, and generating local and specific data.

Materials and methods

San Borja is the capital city of the General José Ballivián Province, located in the department of Beni. It

lies at an altitude of 197 meters above sea level (masl), geographically located 14° 15' 15" South (latitude) and 66° 30' 25" West (longitude). With an annual rainfall of up to 830 mm and an average annual temperature of 26.2° C⁹.

A cross-sectional observational study was carried out, by observing and recording events without intervening in their natural course, the measurement in relation to time was unique¹⁰.

Samples were collected on Fridays and Saturdays in San Borja and processed on Mondays from February to May 2025, at the Veterinary Laboratory Diagnostic and Research Service (SEDILAV) of the Veterinary Medicine and Animal Husbandry Program (CMVZ) of the Faculty of Agronomy at the Universidad Mayor de San Andrés.

The research project was divided into four stages: *Stage 1.* Animal selection, an essential requirement for establishing reference ranges, was carried out by veterinary zootechnicians who certified - apparently healthy animals - through the evaluation of the medical history and body condition 3 (good)¹¹, all those that met the requirements, 173 selected cows entered the study. Sampling was non-probabilistic by convenience because of inclusion criteria, taking into account the study factor: age range and reproductive condition (pregnant and empty). *Stage 2.* Selected animals were fasting on the day of sampling (7 to 9 a.m.). For sample collection, the animal was immobilized in a restraint, 5 mL of blood was taken in syringes from the ventral coccygeal vein, and the samples were transported in a 4° C refrigerator. In the same room in San Borja, the blood serum was separated in a centrifuge at 2500 rpm for 15 min. The samples were then frozen and transported to the laboratory in La Paz. *Step 3.* Once the serum samples

were sent to the SEDILAV laboratory, the records were verified with their respective samples. The STAT FAX 1908 PLUS chemical analyzer was used for serum macromineral analysis. Accuracy quality control was used, with Cromatest Human Multisera Normal and pathological controls, Lot No. 19292 and 19293, respectively. The reagents used were from the same brand. Ca was determined by the end-point colorimetric method, based on the specific binding of cresolphthalein complexone, a metallochromic indicator, and calcium (patient serum) at an alkaline pH, the intensity of the chromophore formed is proportional to the concentration of total Ca¹². Inorganic phosphorus (P) was determined by the end-point ultraviolet method, which reacts with ammonium molybdate in an acidic medium to form a phosphomolybdic complex that is measured at 340 nm¹³. Magnesium (Mg) was carried out by the end-point colorimetric method based on the specific binding of calmagite, a metallochromic indicator, with magnesium at an alkaline pH¹⁴. *Stage 4.* Data analysis, the statistical package Microsoft Excel 2010 and InfoStat, version 2020, were used, and the results were expressed in measures of central tendency (mean, mode and median), standard deviation (SD), coefficient of asymmetry, kurtosis, normal Gaussian bell, analysis of variance (ANVA) and Duncan test, with 95 % reliability.

Results

173 blood serum samples from beef cattle females (SSHBC) were processed; statistical analysis revealed that the values of Ca, Mg and P, the data of central tendency measures (mean, median and mode) were almost similar, the kurtosis and the

asymmetry coefficient were less than $-/+1$, therefore, they have a normal Gaussian bell curve (Table 1).

Table 1 Descriptive statistics of serum magnesium, calcium and phosphorus of beef cattle (*Bos indicus*) San Borja, Beni - Bolivia

Statistical data	Magnesium (mg dL ⁻¹) (n = 173)	Phosphorus (mg dL ⁻¹) (n = 173)	Calcium (mg dL ⁻¹) (n = 173)
Mean	2.40	6.20	9.97
Typical error	.04	.08	.10
median	2.33	6.34	10.00
Mode	2.30	6.30	10.30
Standard deviation	.46	1.02	1.29
Sample variance	.21	1.05	1.66
Kurtosis	-.90	-.58	-.53
Skewness coefficient	.34	-.47	-.09
Mínimum	1.60	3.50	7.20
Máximo	3.40	7.80	12.80
Coefficient of variance	19.16	16.51	12.92
Confidence level (95.0 %)	.07	.15	.19

The data found in the research all fall within a normal Gaussian curve

Table 2 Descriptive statistics and ANVA of serum magnesium (mg dL⁻¹) of female beef cattle (*Bos indicus*) by age range

Statistical data	Magnesium (mg dL ⁻¹)		
	Age Range 1 2 to 4 years n=52	Age Range 2 5 to 7 years n=78	Age Range 3 > to 8 years n=43
Mean	2.45	2.41	2.31
Median	2.35	2.35	2.3
Standard deviation	.47	.46	.44
Mínimum	1.70	1.60	1.70
Máximo	3.30	3.40	3.20
Coefficient of variance	19.15	19.21	18.97

ANVA, $p > 0.05$

Regarding the concentration of Mg in SSHBC by age range (Table 2), in the analysis of variance with a significance level of 0.05 p is equal to 0.3330 ($p > 0.05$), therefore, there is no statistical difference by age range, observing that age range 1 is the one with the highest concentration of magnesium (2.45 mg dL⁻¹).

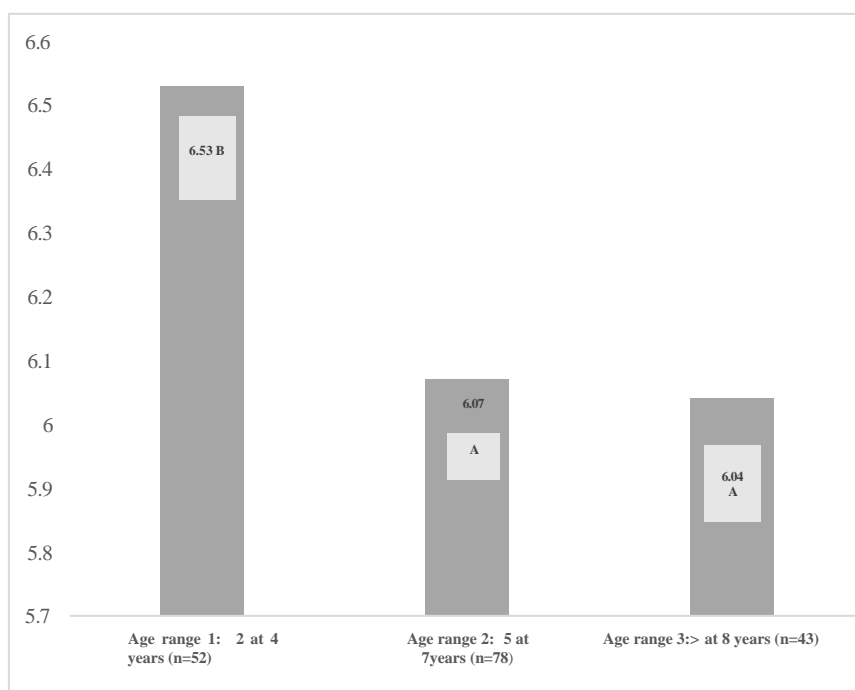
By reproductive condition, pregnant women have a higher concentration of Mg (2.42 mg dL⁻¹), in the analysis of variance with a significance level of 0.05 p is equal to 0.0838 ($p > 0.05$), statistically there is no significant difference by reproductive condition (Table 3).

Table 3 Descriptive statistics and ANVA of serum magnesium (mg dL⁻¹) of beef cattle (*Bos indicus*) by reproductive condition

Statistical data	Magnesium (mg dL ⁻¹)	
	Pregnant condition n=144	Empty condition (n=29)
Mean	2.42	2.26
Median	2.40	2.3
Standard deviation	.46	.43
Mínimum	1.60	1.60
Máximo	3.40	3.30
Coefficient of variance	19.03	18.99

ANVA = $p > 0.05$ **Table 4** Descriptive statistics and ANVA of serum phosphorus (mg dL⁻¹) of female beef cattle (*Bos indicus*) by age range

Statistical data	Phosphorus (mg dL ⁻¹)		
	Age range 1 2 to 4 years (n=52)	Age range 2 5 to 7 years (n=78)	Age range 3 > to 8 years (n=43)
Mean	6.53	6.07	6.04
Median	6.65	5.95	6.3
Standard deviation	.89	1.02	1.11
Mínimum	3.50	3.50	4.00
Máximum	7.70	7.80	7.70
Coefficient of variance	13.64	16.79	18.37

Figure 1 Comparison of Duncan means test for serum phosphorus concentration in (mg dL⁻¹) of beef cattle, according to age range

The concentration of P in SSHBC by age group (Table 4) at analysis of variance was equal to 0.0218 ($p = 0.05$). Statistically, there was a significant difference by age range. Therefore, the Duncan means test (Figure 1) confirmed that age range 1 (2 to 4 years) had the highest phosphorus (6.53 mg dL⁻¹) compared to the other two age ranges. However, age groups 2 (5 to 7 years) and 3 (over 8 years) were statistically equal when comparing means. Pregnant women had

a higher concentration of P (6.25 mg dL⁻¹). The analysis of variance was performed at a significance level of 0.05, with $p = 0.2052$ ($p > 0.05$). There was no significant difference by reproductive status (Table 5). Ca, by age range (Table 6), at the ANVA with a significance level of 0.05 p is equal to 0.5887 ($p > 0.05$), therefore, there is no statistical difference by age range, observing that the age range 1 and 3 the concentration of Ca is 10.08 and 10.03 mg dL⁻¹ re-

spectively. By reproductive condition, the Ca concentration is equal (9.97 mg dL⁻¹), at the ANVA with

a significance level of 0.05 p is equal to 0.9769 (p>0.05), statistically there is no significant difference by reproductive condition (Table 7).

Table 5 Descriptive statistics and ANVA of serum phosphorus (mg dL⁻¹) of beef cattle (*Bos indicus*) by physiological state

Statistical data	Phosphorus (mg dL ⁻¹)	
	Pregnant condition (n=144)	Empty condition (n=29)
Mean	6.25	5.98
Median	6.40	5.8
Standard deviation	1.04	.92
Mínimum	3.50	4.20
Máximo	7.80	7.70
Coefficient of variance	16.64	15.45

ANVA = p>0.05

Table 6 Descriptive statistics and ANVA of serum calcium (mg dL⁻¹) of female beef cattle (*Bos indicus*) by age range

Statistical data	Calcium (mg dL ⁻¹)		
	Age Range 1 2 to 4 years (n=52)	Age Range 2 5 to 7 years (n=78)	Age Range 3 > to 8 years (n=43)
Mean	10.08	9.86	10.03
Median	10.30	9.80	10.00
Standard deviation	1.33	1.27	1.27
Mínimum	7.20	7.20	7.50
Máximo	12.50	12.80	12.40
Coefficient of variance	13.22	12.91	12.7

ANVA = p>0.05 by age range

Table 7 Descriptive statistics and serum calcium (mg dL⁻¹) ANVA of female beef cattle (*Bos indicus*) by reproductive status

Statistical data	Calcium (mg dL ⁻¹)	
	Pregnant condition (n=144)	Empty condition (n=29)
Media	9.97	9.97
Median	10.00	10.10
Standard deviation	1.31	1.20
Minimal	7.20	7.50
Maximum	12.80	12.10
Coefficient of variation	13.13	12.01

ANVA = p>0.05

Table 8 Reference ranges of magnesium, phosphorus and serum calcium of beef cattle (*Bos indicus*) San Borja, Beni Bolivia

	Magnesium (mg dL ⁻¹) p>0.05	Phosphorus (mg dL ⁻¹) p < 0.05		Calcium (mg dL ⁻¹) p > 0.05
		Age Range 2 to 4 years	Age Range ≥ to 5 years	
Media	2.40	6.53	6.06	9.97
D.E.	.46	1.02	1.07	1.29
Media - 2D.E.	1.48	4.49	3.93	7.39
Media + 2D.E.	3.32	8.57	8.19	12.55

Reference Value (Range)	1.5 - 3.3	4.5 - 8.6	3.9 - 8.2	7.4 - 12.6
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Once the measures of central tendency, kurtosis, asymmetry coefficient and ANVA were verified, the reference values were stable with 2 SD with a confidence percentage of 95 % for Mg, a single range of 1.5 to 3.3 mg dL⁻¹ value is declared without considering age range or reproductive condition ($p > 0.05$). In P, 2 range values are declared per age group, ($p < 0.05$) with the Duncan means comparison test, age group 1 is the one with the highest concentration of the mineral, 4.5 to 8.6 mg dL⁻¹ for bovine females from 2 to 4 years old and 3.9 to 8.2 for ≥ 5 years old. For Ca, a single range is declared ($p > 0.05$) from 7.4 to 12.6 mg dL⁻¹ (Table 8).

Discussion

The macromineral values (Mg, P, and Ca) were similar to the measures of central tendency (mean, median, and mode). These values indicate that the data distribution was symmetrical and not biased by extreme outliers. It was observed that most animals had concentrations clustered closely around the central value. The kurtosis and skewness coefficient were less than ± 1 , close to 0, resulting in a near-normal Gaussian curve (Table 1)¹⁵. From a clinical perspective, it allowed for the establishment of reliable reference values (normal range) for the population, significantly benefiting producers, as these data will allow for the optimization of the herd's nutritional, reproductive, and health management.

The concentration of Mg in SSHBC by age range (Table 2) was similar (2.45, 2.41, 2.31 mg dL⁻¹) ($p > 0.05$), data that would fall within the reported reference ranges (1.4 ± 0.44 mmol/L = 3.40 ± 1.07 mg dL⁻¹) for female *B. indicus* cattle in Colombia¹⁶. In a

study conducted on Bolivian Nelore cows (*B. indicus*), values were reported by age group: serum Mg 2 mg dL⁻¹ for multiparous adults aged 10 to 15 years and 1.9 mg dL⁻¹ for young first-time calves aged 3 to 4 years in Santa Cruz, Bolivia¹⁷. It is understood that Mg plays a very important role in neuromuscular transmission and activity, considered a fundamental element for the normal functioning of several enzymatic systems and growth of the bone structure¹⁸.

Thus, the levels of this mineral would cover the needs for vital functions. Calves and calving cows may be more susceptible to Mg deficiency, with requirements varying among calves (6 mg/day), growing animals (30 mg/day), pregnant cows (20 mg/day), and lactating cows (40 mg/day)¹⁹. These requirements would be met according to their nutritional management in the livestock establishment and that was reflected in the results.

We know that Mg is primarily absorbed in the rumen and is decreased by the effect of K and enhanced by Na. Its requirement in calving cows and growing animals must be met directly by the Mg present in pastures²⁰; its serum level was (1.8 to 2.4 mg dL⁻¹)¹⁸. Values below 1.0 to 1.2 mg dL⁻¹ indicate deficiencies, which would be corrected with mineral supplements containing 1 to 3 % Mg²⁰.

The Mg SSHBC concentration by reproductive condition showed no statistical difference ($p > 0.05$) the values in pregnant women were 2.42 mg dL⁻¹ and in empty women 2.26 mg dL⁻¹ (Table 3), values that are similar to those reported by Villa et al¹⁶. Pregnant cows require an increase in Mg. In our study, an average of 2.42 mg dL⁻¹ was observed, favorable for fe-

tal growth and development, so its control and evaluation of its serum concentration is necessary to avoid hypomagnesemia that could occur even with Mg levels close to normal, and is constantly associated with hypocalcemia²⁰.

Within the categories of a bovine herd, the most affected were first-calving females, whose requirements are higher; the data in our work had an average of 2.45 mg dL⁻¹, and the least demanding were multiparous cows older than 8 years, 2.31 mg dL⁻¹. Pregnancy and/or lactation increase the demand for the mineral, increasing the susceptibility of this category to postpartum pathological processes²¹.

P concentrations in SSHBC by age range (Table 4): from 2 to 4 years old, presented values of 6.53 mg dL⁻¹, in the age range 2 and 3, their values were similar (6.07 and 6.04 mg dL⁻¹). In Santa Cruz, Bolivia, P values equal to 5.7 ± 0.1 mg dL⁻¹ were reported in females under 4 years old, and in adult females over 10 years old (5.2 ± 0.1 mg dL⁻¹)¹⁷. It was observed that young animals have higher serum P concentrations, this is because their organisms have not yet experienced continuous pregnancies that could affect these concentrations. Normal P values are considered to be 6 to 8 mg dL⁻¹ in young growing ruminants and 4.5 to 6 mg dL⁻¹ in adult ruminants. Values below 4.5 mg dL⁻¹ are indicative of phosphorus deficiency in cattle²².

Phosphorus is considered the most deficient mineral in grazing cattle. Its deficiency can lead to decreased weight gain, a drop in milk production, and poor reproductive performance, which is easily confused with undernutrition. In severe cases, it also causes alterations in bone structure and teeth (rickets and osteomalacia)²².

Phosphorus is involved in 95 % of energy transfer and utilization reactions, primarily influencing daily

weight gain and is related to the onset of puberty. Furthermore, female gonads are demanding of phosphorus, so deficiency states can delay the onset of ovarian activity, and in extreme cases, deficiency can permanently compromise the function of the reproductive organs²³.

The SSHBC phosphorus concentration by reproductive condition, the results indicate that there is no statistical difference ($p > 0.05$) with values, in pregnant women it is 6.25 mg dL⁻¹ and in empty women it is 5.98 mg dL⁻¹ (Table 5).

Phosphorus requirements for beef cattle vary by age and physiological status, but in general, developing and lactating animals have higher requirements than adult dry cows. For growing and finishing beef cattle, a suggested intake of 0.12 to 0.18 % phosphorus on a dry matter (DM) basis is recommended, while lactating breeding cows may require 0.14 to 0.20 % of DM, and pregnant cows in the last third of gestation may require 0.16 to 0.17 % of DM²².

Ca concentrations in SSHBC by age range and reproductive status (Tables 6 and 7). The results would fall within the values (10.02 ± 0.84 mg dL⁻¹)¹⁶.

Normal concentrations of Ca in apparently healthy cows are usually on average 8 to 10.6 mg dL⁻¹, these ranges may vary by physiological state, age is not a specific factor that determines the concentration, other factors such as growth, pregnancy, and finally calving are the ones that truly influence Ca levels. Growing beef heifers or adult cows need about 6 to 8 g kg⁻¹ of calcium in their diet, although serum values may vary in females already in production or with some higher metabolic need²⁴.

It is important to mention that the bone system constitutes the natural reserve of Ca in living organisms, from these structures it is mobilized all the time, with

a normal serum value of 10 to 12 mg dL⁻¹, regulated by an endocrine system²⁵.

Reference values were determined in SSHBC sampled in San Borja Beni Bolivia for macrominerals with 2 SD: For Mg a single normal range is declared 1.5 to 3.3 mg dL⁻¹, without considering age group and reproductive condition, because statistically there was no significant difference. For P values are declared by age group ($p < 0.05$), young animals (2 to 4 years) 4.5 - 8.6 mg dL⁻¹ and 3.9 - 8.2 mg dL⁻¹ adult animals from 5 to 12 years (Table 8). These values will allow diagnosing subclinical deficiencies, reliable interpretations of macromineral analysis, will help us optimize nutritional management, improve reproductive efficiency and generation of local and specific data.

Consequently, these macrominerals Mg, P and Ca constitute a more precise tool to evaluate the metabolic-mineral status and prevent disorders associated with subclinical alterations in tropical production systems. In addition, this evaluation is a starting point that will allow us to give guidelines to the productive and reproductive performance of *B. indicus*, a species that in the study area is the source of meat and income for the producers in the area.

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Conflicts of interest

This article was prepared and reviewed with the participation of the authors, who declare that there is no conflict of interest that could jeopardize the validity of the results presented.

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Ethical considerations

The animals were restrained and blood was extracted without causing pain or suffering, and was carried out in accordance with current regulations (Animal Protection and Welfare Law, Law 700 of 2015).

Authors' contribution to the article

Marcelina Condori Ticona, developed the research project, analytical processing of blood serum and quality control, blood sample collection, writing, review and editing, interpretation of results, preparation of the original draft, and writing of the article. *Rodrigo Juan Aliaga Alvarez*, developed the selection of apparently healthy animals through clinical diagnosis and auscultation procedures, blood sample collection, writing, review and editing, and interpretation of results, preparation of the original draft, and writing of the article. *Carlos Alejandro Palma Dávila*, developed the selection of apparently healthy animals through clinical diagnosis and auscultation procedures, blood sample collection, writing, review and editing, interpretation of results, preparation of the original draft, and writing of the article.

Limitations in the research

The authors point out that there were no limitations in the present research work.

Access to data

The data and information from this research are present in the article.

Permissions for publication

The authors, once the research was completed, decided by consensus to publish it.

Use of Artificial Intelligence

Initial drafts of the discussion section and bibliographic articles were consulted on ChatGPT, and subsequently reviewed and edited by the authors to ensure accuracy, consistency and appropriateness to the research context.

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